Exogenous Application of 2,4-D, GA₃ and NAA at Flowering Improves Yield and Quality of Kinnow Mandarin (Citrus reticulata Blanco)

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ABSTRACT
This study was undertaken to assess the ability of plant growth regulators (PGRs) to improve fruit set percentage and quality, and to reduce the number of seeds in ‘Kinnow’ mandarin (Citrus reticulata Blanco). Various concentrations of 2,4-dichlorophenoxy acetic acid (2,4-D) (10, 20 and 30 mg/l), gibberellic acid (GA₃) (5, 10 and 20 mg/l) and 1-naphthalene acetic acid (NAA) (5, 10 and 20 mg/l), were applied at the full bloom stage on thirty 10-year-old ‘Kinnow’ mandarin plants grafted onto ‘Rough lemon’ (Citrus jambhiri). The application of 20 mg/l GA₃ significantly reduced the number of seeds/fruit (15.82) compared to the control (19.63). Fruit yield, in terms of number of fruit/tree as well as kg/tree was significantly increased (32.32% in ‘Blood Red’ sweet orange trees treated with 45 mg/l GA₃ compared with the control. Fruit yield, in terms of number of fruit/tree as well as kg/tree was significantly affected by the treatments compared with the control. Similarly, preharvest application of GA₃ can effectively be used to control fruit drop in sweet orange cv. ‘Salustiana’ in which fruit drop percentage was reduced to the lowest value.

INTRODUCTION
Citrus fruits have special importance due to their distinct flavors and therapeutic value. Citrus fruits are an excellent source of vitamin C with fair amounts of vitamins A and B and minerals such as calcium, phosphorus and iron (Nawaz et al. 2008). The juice is refreshing, delicious and soothing. Besides their consumption as fresh fruit, a large number of products and byproducts are prepared and marketed at a premium price (Ahmed et al. 2007a, 2007b). Citrus fruits are ranked at the top in the world and are produced in more than 52 countries around the world. Brazil is the largest producer of citrus worldwide and produces 20.68 million tons (MT) of citrus fruits followed by China, USA, and Mexico with a production of 19.61, 10.01 and 6.85 MT, respectively (Anonymous 2007). Pakistan has a prominent position and is among the top 10 citrus-producing countries in the world. In Pakistan, citrus is cultivated over an area of 185,000 ha with an annual production of 2.2 million ton (Anonymous 2008). Among citrus, ‘Kinnow’ has gained special significance and has monopolized the citrus industry in Pakistan; it is commercially cultivated because of its special significance and has monopolized the citrus industry (Anonymous 2008). Among citrus, ‘Kinnow’ has gained special significance and has monopolized the citrus industry in Pakistan; it is commercially cultivated because of its special significance and has monopolized the citrus industry (Anonymous 2008).

There are other serious problems in Pakistani citrus orchards which are responsible for low yield, low fruit setting and excessive fruit drop, leading to losses for citrus farmers. Low fruit set and reduced quality of fruit is due to malnutrition, water stress, insect pest attack and most importantly, hormonal imbalance (Nawaz et al. 2008). The flowers and fruit of trees senesce when the concentration of auxin decreases and the concentration of abscisic acid (ABA) increases (Browning 1989; Marinho et al. 2005). Endogenous hormones and their balance play a modulating role in the mobilization of nutrients to the developing organs. The application of gibberellic acid (GA₃) increases fruit set (Albrigo and Sauco 2004) and reduces early fruit abscission in parthenocarpic citrus varieties (Fornes et al. 1992).

The use of plant growth regulators (PGRs) has become an important component of agrotechnical procedures. Synthetic auxins have generally been used as an antagonist to reduce abscission of mature fruit with all citrus species but mainly oranges, mandarins and grapefruits (Hield et al. 1964). PGRs play a paramount role in citrus biology and can affect several processes connected with flowering, fruit setting and fruit development. Saleem et al. (2008) reported that final fruit set was significantly affected by GA₃ treatments individually as well as in a mixture with 2,4-dichlorophenoxy acetic acid (2,4-D), with maximum fruit set of 32.32% in ‘Blood Red’ sweet orange trees treated with 45 mg/l GA₃ compared with the control. Fruit yield, in terms of number of fruit/tree as well as kg/tree was significantly affected by the treatments compared with the control. Similarly, preharvest application of GA₃ can effectively be used to control fruit drop in sweet orange cv. ‘Salustiana’ in which fruit drop percentage was reduced to the lowest value.

Keywords: citrus, foliar application, growth regulators, physiochemical properties, seedless
ues (1.70 and 2.28) after the first application of GA3 (100 mg/l), and after the second application of GA3 (50 mg/l) compared to control where fruit drop percentage was 4.50 and 3.85%, respectively (Ibrahim et al. 2011). Although a lot of work has been done to increase fruit set and improve yield and quality in sweet oranges (Citrus sinensis Osbeck), there is no precise recommendation for the control of fruit drop in ‘Kinnow’ mandarin. Moreover, fruit drop of ‘Valencia’ orange in Florida was not reduced by the application of PGRs, stressing the importance of climatic conditions on the effectiveness of PGR treatments (Greenberg et al. 1975). Thus there was a need to test the efficacy of PGRs to increase fruit set, reduce the number of seeds/fruit, improve fruit quality and yield under agro-environmental conditions of Punjab, Pakistan.

MATERIALS AND METHODS

Chemicals and reagents

PGRs, namely 2,4-D, GA3 and 1-naphthaleneacetic acid (NAA) were purchased from BDH Chemicals, Dorset, UK while sodium hydroxide (NaOH), ethanol, Tween-20 and phenolphthalein of reagent grade were purchased from Sigma-Aldrich, St. Louis, MI, USA.

The research work was conducted at the fruit experimental orchard, Institute of Horticultural Sciences, University of Agriculture, Faisalabad, Pakistan. Thirty ‘Kinnow’ mandarin plants grafted onto ‘Rough lemon’ rootstock of uniform size and age (10 years) were selected for this study. Foliar application of PGRs were made at different concentrations [2,4-D (T2 = 10 mg/l, T3 = 20 mg/l, T4 = 30 mg/l); GA3 (T5 = 5 mg/l, T6 = 10 mg/l, T7 = 20 mg/l) and NAA (T8 = 5 mg/l, T9 = 10 mg/l, T10 = 20 mg/l)] during the 2nd week of March at full bloom (3.5 l/tree) to check their effect on fruit set and physiochemical properties of the fruit. T1 was the control, i.e. no PGRs applied. The experiment consisted of 10 treatments (including the control) with three replications; a single tree was considered to be an experimental unit.

Fruit set percentage was recorded from four pre-tagged branches 2.5 cm in diameter, one from each side of the tree in the 2nd week of April. Fruit set percentage was determined by using the following formula (Moeen-Ud-Din et al. 2001):

\[
\text{Fruit set %age} = \frac{\text{Total fruit set}}{\text{Total bloom}} \times 100
\]

Yield/tree was recorded by weighing and counting the total number of fruits/tree at harvest time. Fruit size was measured by randomly measuring the diameter of 10 fruits/tree with the help of a digital vernier caliper. To determine the number of seeds/fruit, 10 fruits were removed from each tree of each treatment; fruit peels were removed manually and the seeds were extracted, counted and the average number of seeds/fruit was calculated.

Peel thickness for each fruit was measured in mm with a digital vernier caliper for the 10 selected fruits from each treatment and then average peel thickness was calculated. The juice of 10 harvested fruit was extracted with the help of manually operated juice squeezer and weighed; average juice weight was calculated separately for each treatment and juice percentage was obtained from the following formula (Nawaz et al. 2008):

\[
\text{Juice %age} = \frac{\text{Average fruit weight}}{\text{Juice weight per fruit}} \times 100
\]

Total soluble solids (TSS) were measured by an automatic digital refractometer (RX 5000, ATAGO, Tokyo, Japan) by placing 1-2 drops of juice on the prism of the refractometer while the acetyl group was determined by adding 1 ml of 2,4-D to each sample with distilled water in a 100 ml beaker; 2-3 drops of phenolphthalein were added for assessing the end point. The samples were titrated against a 0.1N solution of NaOH (Hortwick 1960). The results were expressed as percent citric acid (Ahmed et al. 2006):

\[
\text{Acidity %age} = \frac{N \times 100}{10 \times \text{NaOH used} \times 0.0064}
\]

Vitamin C (ascorbic acid) in juice was estimated according to the method described by Ruck (1961): 10 ml juice was placed into a 250-ml conical flask and the volume was made up to the mark using 0.4% oxalic acid solution. Filtered aliquot (5 ml) was poured into a flask and titrated against 2,6-dichlorophenolindophenol dye until a light pink colour formed which persisted for 10-15 s. Vitamin C content was calculated as:

\[
\text{Vitamin C (mg ascorbic acid/100 ml juice)} = \frac{100}{R_1 \times W \times V_1}
\]

where

\[
R_1 = \text{ml dye used in titration of aliquot;}
V = \text{volume of aliquot made by addition of 0.4% oxalic acid;}
W = \text{volume of aliquot used for titration (ml)}
\]

The PGR solutions for spraying were prepared by dissolving each PGR (2,4-D, GA3 and NAA) in 5 ml of 70% ethanol and then the required solution was prepared by adding distilled water with 0.1% Tween-20 as the surfactant (Khan et al. 2006). Recommended doses of nitrogen (N), phosphorous (P) and potash (NPK) (1000, 500 and 500 g, respectively) fertilizer and 50 kg of farm yard manure (FYM)/plant were supplied to experimental plants (Malik 1994). N was applied in two split doses, first in February in combination with P and K (500g each) while the second application was made in the last week of August (500 g N) and 50 kg of FYM was applied to each plant in December.

The experiment was executed according to a randomized complete block design (RCBD) and the means of data collected were separated by one-way analysis of variance (ANOVA). Significant differences between treatment means were compared by the least significant difference (LSD) test at P = 0.05 (Steel et al. 1996).

RESULTS AND DISCUSSION

There is limited literature regarding the importance of citrus cultivars and PGRs for desert citrus production. PGRs in citrusulture are known to have a profound impact on tree vigour, health, yield and quality of fruit. The results obtained in this study for various parameters are explained next.

Fruit set (%age)

Fruit set is the critical phase in the transformation of a flower to a fruit to obtain good yield and to increase a grower’s returns (Lovatt 1999). Fruit set was counted on marked branches following bloom and petal fall. There were significant differences in fruit set percentage among the various PGR treatments. All PGRs (2,4-D, GA3 and NAA) significantly increased fruit set compared to the control. Maximum fruit set (82.77%) was observed in T3 (5 mg/l GA3) followed by T6 (10 mg/l GA3), T7 (20 mg/l GA3) and T8 (30 mg/l 2,4-D) with fruit set being 77.80, 76.63 and 63.35%, respectively. The lowest fruit set (43.89%) was observed in T1 (control). 2,4-D and NAA at high concentrations increased fruit set less efficiently, i.e., T9 (20 mg/l NAA) and T10 (30 mg/l 2,4-D) compared to lower concentrations, i.e., T5 (10 mg/l 2,4-D) and T8 (5 mg/l NAA). Our results regarding fruit set were in agreement with those of Huang and Huang (2005), who concluded that a foliar spray of GA3 at 50 mg/l on % flower fall stage of citrus resulted in good protection of fruitlets and increased fruit set and yield by 100% compared to the control in ‘Nanfengmiju’ mandarin. Similarly, Agusti et al. (1982) found that a single spray of GA3 (5 mg/l) at the petal fall stage increased fruit set significantly (29%) in ‘Navelate’ sweet orange. The application 2,4-D (100 mg/l) and GA3 (100 mg/l) to flowering branches increased fruit set by 17.3 and 21.4%, respectively in Ginkgo (Ginkgo biloba) trees (Zhao et al. 2005). The results of our study indicate that 5 mg/l GA3 can be used to enhance fruit set in ‘Kinnow’ mandarin.
Yield

Fruit yield is of prime concern to orchardists. They grow plants for better yield and good quality fruit production (Ahmed et al. 2006). Yield is measured in terms of number of fruits/plant and fruit weight/plant; the results regarding both of these parameters are described next.

1. Number of fruits/plant

There were significant differences among various treatments. All PGR treatments increased the total number of fruits/plant (Table 1). The highest number of fruits/plant (682.33) was observed in T5 (5 mg/l GA3) followed by T2 (10 mg/l 2,4-D) and T5 (5 mg/l NAA) with 659.66 and 647.66 fruits/plant, respectively; the lowest number of fruits/plant (422.00) was found in T1 (control). All three PGRs, at lower concentrations, increased yield but at higher concentrations fewer fruits/plant were observed in most treatments. Since 2,4-D and NAA (auxin) have a dual function, i.e., when they are used at lower concentrations they act as growth promoters while at higher concentrations they act as growth inhibitors, they sometimes adversely affect the vegetative and reproductive behaviour of citrus (Saleem et al. 2008). This might explain why fewer fruits/plant formed at higher concentrations of GA3 by promoting vegetative growth at the expense of reproductive growth (fruits). The intensity of leaves on a flush was significantly affected by PGR treatments compared to the control in ‘Blood Red’ sweet orange. A maximum number of leaves (13.76) formed on trees treated with 30 mg/l of a mixture of GA3 + 2,4-D each, while fewest leaves formed in control trees having 4.11 leaves/flush; similarly, shoot length and maximum shoot length (7.39 cm) also significantly increased in trees treated with GA3 + 2,4-D but the minimum size of a flush (2.30 cm) was recorded in control trees (Saleem et al. 2008).

2. Fruit weight/plant (kg)

Maximum fruit weight/plant (96.14 kg) was observed in T5 (5 mg/l GA3) closely followed by T2 (10 mg/l 2,4-D) and T5 (20 mg/l GA3) with fruit weight/plant being 90.14 and 89.17 kg, respectively. Minimum fruit weight/plant (52.49 kg) was observed in T1 (control). In this case it is clear from the data that higher yield was observed at higher concentrations of PGRs, except for GA3 (Table 1); at higher concentrations of GA3 fewer fruits/plant were observed while fruit size was greater at higher concentrations; consequently, greater fruit weight/plant was observed. T5 (5 mg/l GA3) and T2 (10 mg/l 2,4-D) were the best treatments. The results for yield were similar to those of Agusti et al. (1982), who reported that the application of 5% fruit weight of GA3 on ‘Navelate’ sweet orange at the end of petal fall stage significantly increased yield up to 89 kg/tree compared to untreated trees (80 kg/tree). Huang and Huang (2005) reported that spraying GA3 (50 mg/l) on citrus achieved good results by protecting fruitlets and increasing yield in ‘Nanfengmiju’ mandarin. Similarly, Saleem et al. (2008) observed that application of 45 mg/l GA3 to 15-years-old ‘Blood Red’ sweet orange plants at the full bloom stage increased yield (71 kg/tree) more than the control (48 kg/tree).

Fruit size (mm)

The size of the fruit is important, not only because it is a component of productive yield, but also because it determines the acceptance of fruit by the consumer. The importance of fruit size as a parameter of quality of citrus fruits has increased markedly in recent years. The consumer’s preference for large fruit causes huge differences in price between large and small fruit to the point that income from smaller fruit is often less than the picking and handling costs (Guardiola and Garcia-Luis 2000).

The exogenous application of PGRs significantly increased the fruit size of treated plants compared to the control (Table 1). However, 2,4-D and GA3 increased fruit size more. The fruit with greatest diameter (73.63 mm) was found in T6 (10 mg/l GA3) followed by T2 (20 mg/l 2,4-D; 72.57 mm); smallest fruit (64.02 mm diameter) was found in T1 (control). All three PGRs increased fruit size more than the control, with the best PGR treatments being 10 mg/l GA3 (T3) and 20 mg/l 2,4-D (T5). Our results agree with those of Thomas and Lovatt (2004), who reported that foliar application of 15 mg/l 3,5,6-trichloro-pirydioxacetic acid (3,5,6-TPA) increased fruit size most in ‘Clementine’ mandarin and ultimately increased yield due to larger fruits. The net increase in yield/acre averaged over the 3-year experiment for the 3,5,6-TPA-treated trees, compared to the control trees, was: 610.00 kg of jumbo (fruit 63.51-69.85 mm in diameter); 1289.56 kg of large (57.16-63.50 mm); and 1165.73 kg of medium (50.81-57.15 mm) size fruit. No other treatment increased the yield of large size fruit compared to the control. Similarly, Zhang and Whiting (2011) noted that 200 mg/l GA3 applied at 9 days after full bloom improved final fruit (‘Bing’ sweet cherry under USA field conditions) weight by 15%, and 56% of the fruit from this treatment had fruit weight/fruit > 9 g; to the weight of untreated limbs only increased 15%.

Percentage juice

Juice is an extremely important parameter for industrial processing, also related to size, which, in turn, although determined by the genetic characteristics of a cultivar, is affected by cultural practices such as the application of PGRs (Nawaz et al. 2008). Significant differences in juice percentage, calculated on the basis of juice weight, were observed among the various treatments (Table 1). The application of PGRs significantly increased the juice percentage, maximum (53.29%) in T5 (10 mg/l GA3) followed by T7 (20 mg/l GA3) and T4 (30 mg/l 2,4-D) with 51.18 and 50.76%, respectively; the lowest juice percentage (42.56%) was found in T1 (control). The juice percentage could be increased by as much as 10% simply by the application of PGRs. An increase in juice percentage was observed following the application of PGRs, which affect many physiological and biochemical processes within plants. Davies et al. (1997) also concluded that the application of GA3 (18 g ai (active ingredient)/acre) had the potential to improve processing juice extraction weight by 3.2 to 9.4% depending on the cultivar and harvesting time.

Peel thickness (mm)

Peel thickness is an important parameter of fruit quality. There were no significant effects of PGRs on peel thickness (Table 1) although maximum peel thickness (3.48 mm) was observed following the application of GA3 at 20 mg/l (T5), least in T2 (10 mg/l 2,4-D). Agusti et al. (1994) reported that the application of 50 mg/l 2,4-DP (2,4-dichlorophenoxypropionic acid) applied at the end of physiological drop on ‘Owari Satsuma’ mandarin (Citrus unshiu Marc.) increased fruit size and juice percentage significantly while no significant effect on peel thickness was observed as the peel thickness of treated fruit was 3.74 mm compared to the control (3.66 mm); however, a denser and firmer peel was produced. Similarly, Nawaz (2007) reported that preharvest application of PGRs [2,4-D (10, 20, 30 mg/l), GA3 (10, 50, 100 mg/l) and NAA (10, 15, 20 mg/l) at the preharvest fruit drop stage had no significant effect on peel thickness in ‘Kinnon’ mandarin.

Number of seeds/fruit

Seed dispersal in fleshy fruits like citrus is an unwanted characteristic. ‘Kinnon’ fruit should be seedless – or at least with less seed/fruit – because seeded fruits are more inconvenient to eat and add bitterness during processing, thereby satisfying the demands of customers and the processing industry (Ahmed 2005). The data regarding plump seed
The treatments clearly formed three groups (Table 1): auxins (T2 (10 mg/l 2,4-D), T3 (20 mg/l 2,4-D), T4 (30 mg/l 2,4-D)), NAA (T5 (5 mg/l GA3), T6 (10 mg/l GA3), T7 (20 mg/l GA3), T8 (5 mg/l NAA), T9 (10 mg/l NAA), T10 (20 mg/l NAA)) formed the 1st group with a high number of seeds; GA3 (T6 (10 mg/l GA3) and T7 (20 mg/l GA3)) formed the 2nd group with relatively fewer seeds; T2 (10 mg/l 2,4-D) formed the 3rd group with the fewest seeds/fruit. GA3 reduced the number of seeds compared to the control and other treatments. The reduction in the number of seeds/fruit may be due to the stimulatory effect of GA3 on parthenocarpic fruit development (Zhang 2003). It can be inferred from the data that PGRs can reduce the number of seeds in ‘Kinnow’ mandarin, which is highly prized but contains many seeds which is discouraging for the citrus juice industry/producers. However, to reduce the number of seed, further detailed studies are required to standardize the concentration and time of application with special reference to our local agro-ecological conditions.

Chemical characteristics

1. Total soluble solids (TSS)

TSS is an important measure of the sugar content of fruits as sugars constitute approximately 85% of the soluble solids in citrus fruits (sweet oranges and mandarins) (Waradowski et al. 1979). Maximum TSS (11.73%) accumulated in T9 (10 mg/l NAA) followed by T8 (5 mg/l NAA) and T2 (30 mg/l 2,4-D) i.e., 11.38 and 11.37%, respectively; minimum TSS (9.67%) was observed in T1 (control). All the three PGRs increased the TSS compared to the control. It can also be inferred from the data that auxin (2,4-D and NAA) increased TSS more efficiently than GA3. Ingle et al. (2001) reported that foliar sprays of 10 mg/l 2,4-D, 30 mg/l NAA and 25 mg/l GA3 in February, August and September significantly increased fruit quality parameters such as juice percentage, TSS, ascorbic acid content, acidity and peel thickness, similar to our findings. Similarly, Nawaz et al. (2008) reported a significant increase in TSS compared to the control in ‘Kinnow’ mandarin following the foliar application of various concentrations of 2,4-D at the preharvest stage; 30 mg/l 2,4-D increased TSS to 12.03% compared to the control which had a TSS of 10.43%. However, Berhow (2000) found that early foliar spray treatments of 100 mg/l GA3 on grapefruit (Citrus paradisii Macf.) significantly lowered the concentration of the bitter flavonoid naringin in grapefruit fruit tissues and finally resulted in larger mature fruit, which yielded juice with the same TSS value as juice obtained from control fruits.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Fruit set (%)</th>
<th>Number of fruits/plant</th>
<th>Fruit weight/ plant (kg)</th>
<th>Fruit size (mm)</th>
<th>Juice (%)</th>
<th>Peel thickness</th>
<th>Number of seeds/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1 (control)</td>
<td>43.89 e</td>
<td>422.00 c</td>
<td>52.49 d</td>
<td>64.02 c</td>
<td>42.56 c</td>
<td>3.15</td>
<td>19.63 a</td>
</tr>
<tr>
<td>T2 (10 mg/12,4-D)</td>
<td>63.35 bc</td>
<td>659.66 ab</td>
<td>90.96 b</td>
<td>68.90 b</td>
<td>50.09 ab</td>
<td>3.00</td>
<td>18.60 ab</td>
</tr>
<tr>
<td>T3 (20 mg/12,4-D)</td>
<td>55.17 cd</td>
<td>644.66 ab</td>
<td>88.63 ab</td>
<td>72.57 ab</td>
<td>48.70 b</td>
<td>3.06</td>
<td>19.40 a</td>
</tr>
<tr>
<td>T4 (30 mg/12,4-D)</td>
<td>51.23 cd</td>
<td>610.00 ab</td>
<td>89.13 ab</td>
<td>72.16 ab</td>
<td>50.76 ab</td>
<td>3.11</td>
<td>18.20 abc</td>
</tr>
<tr>
<td>T5 (5 mg/l GA3)</td>
<td>82.77 a</td>
<td>682.33 a</td>
<td>96.14 a</td>
<td>72.36 ab</td>
<td>49.59 ab</td>
<td>3.21</td>
<td>16.65 bc</td>
</tr>
<tr>
<td>T8 (5 mg/l NAA)</td>
<td>77.80 ab</td>
<td>631.33 ab</td>
<td>85.92 ab</td>
<td>73.63 a</td>
<td>53.29 a</td>
<td>3.31</td>
<td>16.18 cd</td>
</tr>
<tr>
<td>T3 (20 mg/l NAA)</td>
<td>76.63 ab</td>
<td>624.00 ab</td>
<td>89.17 ab</td>
<td>70.36 ab</td>
<td>51.18 ab</td>
<td>3.48</td>
<td>15.82 a</td>
</tr>
<tr>
<td>T7 (20 mg/l GA3)</td>
<td>60.33 bc</td>
<td>647.66 ab</td>
<td>86.50 ab</td>
<td>66.90 ab</td>
<td>49.49 ab</td>
<td>3.34</td>
<td>19.40 a</td>
</tr>
<tr>
<td>T1 (control)</td>
<td>52.79 cd</td>
<td>610.33 ab</td>
<td>79.64 c</td>
<td>69.90 ab</td>
<td>49.04 b</td>
<td>3.30</td>
<td>19.22 a</td>
</tr>
<tr>
<td>T5 (5 mg/l NAA)</td>
<td>53.04 cd</td>
<td>642.66 ab</td>
<td>87.46 ab</td>
<td>69.23 ab</td>
<td>50.73 ab</td>
<td>3.26</td>
<td>18.87 a</td>
</tr>
<tr>
<td>T6 (10 mg/l GA3)</td>
<td>50.79 cd</td>
<td>610.33 ab</td>
<td>79.64 c</td>
<td>69.90 ab</td>
<td>49.04 b</td>
<td>3.30</td>
<td>19.22 a</td>
</tr>
</tbody>
</table>

Table 1 Effect of growth regulators on chemical characteristics of ‘Kinnow’ mandarin.

Means in a column followed by similar letters are not significantly different at P = 0.05 (LSD).

2. Percentage acidity

Fruits sprayed with PGRs showed a significant decrease in acidity. Highest acidity (1.10%) was recorded for T1 (control) followed by T2 (10 mg/l 2,4-D) i.e., 1.01% whereas, minimum acidity (0.78%) was recorded in T10 (20 mg/l NAA). For all other treatments it ranged between 0.97 and 0.90%. The application of PGRs reduced the acidity of fruit which is a desirable character for superior fruit quality. Fruit harvest in ‘Kinnow’ is delayed in Pakistan due to high acidity of juice which imparts a bitter taste. Harvesting can be hastened by at least 15 days by using 15 mg/l NAA at the preharvest stage (Nawaz et al. 2008).

Vitamin C

Vitamin C (ascorbic acid) is essential for life and is a powerful water-soluble antioxidant, it must be ingested for survival as it prevents humans from many serious diseases like atherosclerosis and cancer by scavenging reactive oxygen species (ROS) and protecting tissues from ROS-induced oxidative damage (Parke 1990; Padayatty et al. 2003). The concentration of vitamin C content in fruits varies for various citrus species and is affected by environmental factors, the time of fruit harvest, plant vigour, age of the plant and by the application of PGRs (Ingle et al. 2001). Vitamin C content was significantly increased by various concentrations of PGRs, maximum (45.11 mg/100 ml) in T9 (10 mg/l NAA) and T7 (20 mg/l GA3) 0.97 mg/l GA3. Minimum ascorbic acid content (35.84 mg/100 ml) was observed in T1 (control). The application of auxins (2,4-D and NAA) was better than the use of GA3 in improving the vitamin C content of ‘Kinnow’ mandarin (Table 2).

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Total soluble solids (TSS)</th>
<th>Acidity (%)</th>
<th>Vitamin C (mg/100 g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1 (control)</td>
<td>9.67 b</td>
<td>1.01 a</td>
<td>35.84 d</td>
</tr>
<tr>
<td>T2 (10 mg/12,4-D)</td>
<td>11.34 a</td>
<td>0.94 a</td>
<td>39.32 bcd</td>
</tr>
<tr>
<td>T3 (20 mg/12,4-D)</td>
<td>10.97 a</td>
<td>0.91 b</td>
<td>43.28 abc</td>
</tr>
<tr>
<td>T4 (30 mg/12,4-D)</td>
<td>11.37 a</td>
<td>0.91 b</td>
<td>41.96 abc</td>
</tr>
<tr>
<td>T5 (5 mg/l GA3)</td>
<td>10.95 a</td>
<td>0.91 b</td>
<td>39.10 bcd</td>
</tr>
<tr>
<td>T6 (10 mg/l GA3)</td>
<td>11.13 a</td>
<td>0.94 b</td>
<td>38.73 cd</td>
</tr>
<tr>
<td>T7 (20 mg/l GA3)</td>
<td>10.93 a</td>
<td>0.97 b</td>
<td>41.91 abc</td>
</tr>
<tr>
<td>T8 (5 mg/l NAA)</td>
<td>11.30 a</td>
<td>0.90 b</td>
<td>41.02 abc</td>
</tr>
<tr>
<td>T9 (10 mg/l NAA)</td>
<td>11.73 a</td>
<td>0.94 b</td>
<td>45.11 a</td>
</tr>
<tr>
<td>T10 (20 mg/l NAA)</td>
<td>11.38 a</td>
<td>0.78 c</td>
<td>43.80 ab</td>
</tr>
</tbody>
</table>

Means in a column followed by similar letters are not significantly different at P = 0.05 (LSD).

CONCLUSION

PGRs are versatile compounds. Exogenous application of PGRs (2,4-D, GA3, NAA) in this study significantly increased ‘Kinnow’ mandarin fruit set percentage and increased the average fruit yield/plant. Fruit quality (juice percentage, TSS, acidity and fruit size) was also improved.
by the application of PGRs which, at lower concentrations, enhanced fruit parameters more efficiently than higher concentrations. Of particular interest, GA\textsubscript{3} significantly reduced the number of seeds/fruit compared to the control. However, more detailed studies are required to check the efficacy of GA\textsubscript{3} in producing seedless/ or less seeded fruits in ‘Kinnow’ and other citrus cultivars of commercial importance.

REFERENCES


Davis FS, Campbell CA, Zelman G (1997) Gibberellic acid sprays for improving fruit peel quality and increasing juice yield of processing oranges. *Proceedings of the Florida State Horticultural Society* 110, 16-21


Held HZ, Burn RM, Coggins CW (1964) Preharvest use of 2,4-D on Citrus, California Agriculture and Experimental Station, Circular, 10 pp


Huang JH, Huang L (2005) The application of GA\textsubscript{3} in citrus orchards. *South China Fruits* 3, 32-36


Ruck JA (1961) *Chemical Method for Analysis of Fruit and Vegetable Products*, Research Station Summerland, Research Branch, Chanda, Department of Agriculture No 1154


Thomas CC, Lovatt C (2004) Application of *Plant Growth Regulators and/or Fertilizers to Increase Fruit Set Fruit Size and Yield of Clementine Mandarin, Citrus Research Board*, Annual Report, Botany and Plant Sciences, University of California, Riverside, USA, pp 1-3


Zhang GX (2003) Effects of GA\textsubscript{3} and CPPU on inducing seedless fruit and fruit quality of Bendizao mandarin variety. *South China Fruits* 32, 8-10

Zhao Y-J, Bian X-B, Tao J (2005) Effects of GA\textsubscript{3} and 2,4-D on fruit set of ginkgo. *South China Fruits* 2, 54-58